Revolutionizing Turkey Production

Functional genomics is the driving force

onsumers give many thanks for the estimated 5.6 billion pounds of turkey brought to market each year. U.S. farmers meet that demand by producing close to 300 million turkeys annually.

For decades, turkey breeders have produced bigger, meatier birds by using advanced genetic selection. The resulting difference in size between males, or toms, and females, or hens, requires producers to rely solely on artificial insemination for reproduction. It's a labor-intensive and tricky process.

Because an average turkey farm has about 33,000 sexually mature hens to inseminate, scientific advancements are in demand.

Scientists at the ARS Biotechnology and Germplasm Laboratory in Beltsville, Maryland, are using functional genomics to improve reproductive methods used by turkey farmers. Functional genomics is the study of the function of genes in a particular cell and how these

The researchers are focusing on differences in gene expression within cells in a special site inside the hen's reproductive tract, called the oviductal sperm-storage tubules (SSTs).

Turkey hens are able to store and keep sperm alive in their SSTs for up to 75 days. But sperm collected for artificial insemination—through a process called "milking"—can stay alive for only 8 to 10 hours outside SSTs. And that's after they're protected in vials and buoyed by a mixture of nutrients and other fluids called "extenders."

Such a narrow window of opportunity in which to use or lose the sperm poses difficult time limits for farmers.

A SAGE Approach

The researchers are delving into the mystery of how sperm stay healthy for so long inside a hen's reproductive tract. This information could help improve the capability of keeping sperm alive outside the SSTs after milking.

genomics. It's called SAGE (Serial Analysis of Gene Expression), and Zuelke has since found it to be spectacularly effective for researching livestock genomics.

"In terms of today's genetics research, functional genomics is where the rubber meets the road," says Zuelke. His team is using SAGE to study alterations in gene expression when turkey sperm are present in SSTs.

Expressed genes are those genes that are "switched on"—that is, making important proteins—at a given time. Using the SAGE method, the researchers can count the number of RNA transcripts, or copies, made by expressed genes. Many transcripts signal high expression; few transcripts suggest low expression.

"The beauty of SAGE is that it counts close to exactly the number of transcripts of every expressed gene in the entire turkey genome at a particular moment by using digital technology," says Zuelke. "If you have 20 transcripts, then in theory you have 20 SAGE tags."



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Improving Sperm Preservation

To maintain a fertility rate of 90 percent or more on turkey farms, each hen is inseminated once a week—with about 200 to 300 million sperm each time—for 5 months. Each collected dose of turkey semen yields about 9 billion to 12 billion sperm per milliliter.

"Lengthening the time that these sperm stay alive outside the tom would translate into significant savings for the commercial turkey industry," says Zuelke.



At the Biotechnology and Germplasm laboratory in Beltsville, Maryland, physiologists Julie Long and Kurt Zuelke evaluate whether turkey sperm are alive or dead in studies to improve the lifespan of stored turkey sperm.



Physiologist Kurt Zuelke (left) and biologist Wes Garrett select turkey sperm storage tubule proteins from a two-dimensional gel for analysis by mass spectrometry as a followup to SAGE analysis.

The first breakthrough study was conducted by Zuelke, reproductive physiologist Julie Long, and ARS researchers at the Bovine Functional Genomics Laboratory, also in Beltsville.

The scientists ran two SAGE analyses—one on tissue taken from turkey SSTs when sperm were present and one when sperm were not present. They reported that more than 214 genes were expressed at different levels within SSTs when sperm were present compared to when sperm were not present.

This was the first report in scientific literature that indicated that the presence or absence of sperm in the reproductive tract could affect gene expression within the SSTs. The breakthrough lays a foundation for understanding which proteins are produced from the expressed genes.

"SAGE technology allows us to study thousands of genes simultaneously, measure their expression, and quickly identify genetic differences between cells in the presence and absence of sperm," says Zuelke.

During the study, the team found a particular gene, named "avidin," which corresponded to a SAGE tag. Avidin had already

been identified and well studied in chickens. Since the SAGE technique had never been used in poultry before, *avidin* was chosen to help validate SAGE in turkeys. "Now that we have the *avidin* gene, we can use it as a model for analyzing other genes expressed in the presence of sperm," he says.

Learning which genes are switched on when sperm are present in the SSTs can help identify which proteins are produced under those special circumstances. Those proteins could then be put into extenders to help prolong the life of sperm outside the SSTs.

Future Applications

The scientists consider the milestone of identifying key expressed genes as opening a door to further research. They hope that their discoveries will lead to further knowledge that will eventually allow them to freeze turkey sperm.

"If we could extend their survival rate to 24 hours, allowing them to stay viable overnight, that would open wider transportation and storage options for better reproductive management," says Zuelke.

The newly developed turkey SAGE tag data has been donated to a database called SAGE-Map, which is maintained by the National Center for Biotechnology Information, part of the National Institutes of Health, in Bethesda, Maryland. SAGE-Map is available to both public and private scientists for further research and can be accessed at www.ncbi.nih. gov.—By Rosalie Marion Bliss, ARS.

This research is part of Food Animal Production, an ARS National Program (#101) described on the World Wide Web at www.nps.ars.usda.gov.

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